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COMPLETE SPECIFICATION

Therapeutic Bone Mixture

We, OLIN MATHIESON CHEMICAL CORPORATION, a corporation organised under the laws of the State of Virginia, United States of America, of Ten Light Street, Baltimore 3, Maryland, United States of America, do hereby declare the invention, for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement:—

The present invention relates to the treatment of non-human animal bones for transplantation without deleterious effects at later times to other living animal bodies including human beings.

A method is known of preserving and categorizing human bones for transplantation. Since the supply of suitable bones which may be preserved in accordance with the known method is limited, it is desirable to provide a method of preserving bones from other sources for transplantation to humans without deleterious effects.

The present invention provides a bone mixture comprising pulverized non-human animal bone that has been cultured in a refrigerated state in a bath of an antibacterial or antifungal agent and a blood component from the same species animal from which the bone was derived, and plasma clot substance as a binder for said pulverized bone, said plasma clot substance being derived from blood plasma obtained from the same species animal as the bone.

The invention also includes a method of preparing such a bone mixture, which comprises maintaining non-human animal bone in a refrigerated state in a bath containing an antibacterial or antifungal agent and a blood component from the same species animal from which the bone was derived, pulverizing the bone thus maintained, and blending the pulverized bone with plasma clot substance as a binder, said plasma clot substance being derived from blood plasma obtained from the same species animal as the bone.

Application No. 24809/56 (Serial 827,746) described and claims a process of preserving non-human animal bones for subsequent transplantation to other living animal bodies including human beings simultaneously encouraging cell growth during such preservation, which comprises removing bone from a healthy non-human animal body source under sterile conditions, and placing the removed bone in a container having therein a systemic antibacterial agent and blood plasma or blood serum from the same specie animal from which the bone was removed.

Bones from any suitable non-human source may be used in practicing such process including, by way of example, domestic animals such as sheep, cattle, pigs, dogs, cats and horses. However, regardless of the source, the bone must be removed from the non-human animal body source under sterile conditions so as to inhibit contamination thereof by foreign matter or bodies which might cause harmful effects during preservation of the bone or upon transplanting the preserved bone to another body. Also, it is desirable that the bone be obtained promptly after death of the animal while the bone is still alive in order to inhibit necrosis and atrophy. Also, it is desirable that only healthy bone, free from disease, be preserved in order to prevent obvious harmful effects to the bone during preservation or to the bone bed to which such preserved bone may be transplanted; therefore, the animals should be tested to eliminate those with disease.

It is not necessary to classify the various animal bones as to types, nor is it necessary in practicing such process to classify the blood of the original source as to type or characteristic. However, in order to preserve the bone and to encourage and maintain new cell growth during preservation of the bone, it is necessary that the bone be maintained in blood plasma or blood serum which must be from the same species animal as that of the bone being preserved.

When the bone is removed from the animal source and placed in a container with blood

[*Price 3s. 6d.*]

plasma or blood serum from the same species animal as the bone being preserved therein, new cell growth occurs during the period of preservation so long as the blood plasma or blood serum furnishes proper nutrition for the bone and the new cells.

Microscopic examinations of portions of the stored bone may be made at suitable intervals to determine if the bone cells have begun to atrophy or if necrosis has occurred. Both these conditions, if noted upon examination, may be overcome, and proper growth and life conditions of the bone may be maintained by replenishing the container with a new and fresh supply of blood plasma or blood serum. The blood plasma or blood serum added must, of course, be from the same species animal as the bone being preserved. To this extent it is necessary that proper indication of the kind of bone in each container be noted at the time that such bone is removed from the source and placed in the container.

Bone, when removed from the body source, is placed in a container of a suitable type such as glass and of a suitable size to accommodate the bone being preserved. However, any suitable storage material which is inert relative to the contents contained therein and which will not deteriorate upon storage may be used.

The fluid to be placed in the container may consist of blood plasma along with any suitable antibacterial or antifungal agent such as penicillin or sodium sulfadiazine. The blood plasma used may be diluted up to 90 percent by volume with normal saline solution; however, since the growth of new cells and the maintenance of bone life is dependent upon the presence of sufficient blood plasma, a stronger solution may be used if the bone is to be stored indefinitely. Also, use of a stronger solution such as, for example, whole blood plasma will eliminate the necessity of making frequent examinations of the bone to determine if necrosis or atrophy has occurred. Blood serum may be used; however, the bone will not be maintained viable as long as it would if blood plasma is used.

If the container contents are maintained at relatively low temperatures as compared with the normal body temperature of the body source of the bone, the rate of depletion of nutrition from the blood plasma or blood serum to the bone is materially reduced. However, if the temperature is lowered so that the container contents are frozen, the bone being preserved dies and will not become viable upon transplantation to a living body. Therefore, in order to reduce the intervals at which it may be necessary to replenish the blood plasma or blood serum in the container on the one hand, while on the other hand maintaining the bone being preserved viable while simultaneously encouraging fibroblastic cell growth of the preserved bone, the storage temperature should be maintained at about 50° C. Higher temperatures may be used without harming bone life or cell growth.

All non-human animal bones, when preserved by this process, not only are maintained viable, but a fibroblastic cell growth is encouraged on the bone. The fibroblastic cell is the most primitive of bone cells, and its growth is encouraged and maintained by the blood plasma or blood serum in the container. The blood plasma also helps the growth of periosteum and apparently converts the osteocyte to fibroblastic tissue.

The blood plasma or blood serum used should be from the same specifies animal as the bone being preserved. It is not necessary to provide blood plasma or blood serum from the same animal which served as the bone source. For example, if oxen bone is being preserved, then any oxen blood plasma or blood serum may be used as the preserving medium; if horse bone is being preserved, then any horse blood plasma or blood serum may be used. Oxen blood plasma cannot be used for successfully preserving a bone of an animal of a different species.

In accordance with the present invention, non-human animal bone so preserved is removed from the organic liquid and is treated as by sawing it into small pieces and then pulverizing the pieces as in a grinder or mixer, it being necessary to regulate the speed of the grinder or mixer so as not to cauterize the bone and thereby hamper its properties for projected uses as in surgical and dental operations.

The pulverized bone is then mixed with a binder which is pooled homologous plasma clot substance, for example, blood plasma from the same species animal supplying the bone and the mixture is cooled at about 40° F. The clot results from the reaction between the prothrombin and thromboplastin from the blood plasma and the calcium from the bone to form thrombin, which in turn reacts with the fibrinogen of the blood plasma to form a fibrin clot. In mixing the blood plasma and pulverized bone, careful sterile technique must be employed.

As to proportions, it has been found that a suitable gel of pulverized bone and plasma clot substance can be made when the ratio of bone to plasma clot approximates 10 to 1; but other ratios serve the purpose as well, it being necessary that the plasma clot binder is present in such proportions as to ensure a gel or binding between the bone particles. For the purpose of obtaining a homogenized mixture, any suitable blender or mixer may be employed.

After mixing, if the compound or paste thus formed is not used, it must be returned immediately to a refrigerated state and maintained therein under sterile conditions, a range of refrigerating temperatures from 40°

F. to 50° F. serving to maintain the refrigerated state but successful refrigeration being also maintainable outside of this range limit. As may be required, additional blood plasma may in time be added to sustain the paste; and one example of accomplishing this would be enclosing the paste in an osmatic membrane which the blood plasma surrounds. In this regard, sustaining the paste means replenishing the blood plasma level in a container in cases where the plasma level therein might otherwise have been dimished to stand below a portion of the paste. When an osmotic membrane is employed, as aforesaid, the membrane surrounds the paste to hold it in form, and the blood plasma level must thus be maintained within the container to stand at a level above the uppermost part of the membrane.

When incisions are made, as in the case of preparing a patient to receive bone preserved as described above for splints, bridging elements, or grafts, the introduction into the incision of the present paste or admixture of ground or pulverized bone will result in an acceleration of the clotting action since the organic calcium and phosphorus of the mixture combine in such presence with any uncombined portion of the plasma clot and also combine with constituents of the blood from the incision to form blood clots to lessen the bleeding from the incision.

The present bone mixture also can serve as a bed or filler between bone sections as, for example, when such sections are being grafted. The viable paste will first grow fibroblastic cells which cells in turn develop into obsteoblastic cells and serve, as does the whole bone preserved as described above, as a bridging element and conduit for nutrients until such time as replacement occurs in the course of metabolic action.

The present bone mixture also has adaptability in dentistry as a filler for dental cavities as, for instance, when the extraction of a tooth may be accompanied by the removal of parts of the jawbone. In such case the bone mixture bridges the cavity; and its growth of fibroblastic cells which develop into osteoblastic cells cooperates with the similar growth from the jawbone or molar parts until such time as replacement occurs in the due course of metabolism.

WHAT WE CLAIM IS:—

1. A bone mixture comprising pulverized non-human animal bone that has been cultured in a refrigerated state in a bath of an antibacterial or antifungal ugent and a blood component from the same species animal from which the bone was derived, and plasma clot substance as a binder for said pulverized bone, said plasma clot substance being derived from blood plasma obtained from the same species animal as the bone.

2. A bone mixture according to claim 1, in which the blood component is blood plasma or blood serum.

3. A bone mixture substantially as hereinbefore described.

4. A method of preparing a bone mixture, which comprises maintaining non-human animal bone in a refrigerated state in a bath containing an antibacterial or antifungal agent and a blood component from the same species animal from which the bone was derived removing the bone from the organic liquid, pulverizing the bone, and blending the pulverized bone with plasma clot substance as a binder, said plasma clot substance being derived from blood plasma obtained from the same species animal as the bone.

5. A method according to claim 4, in which the blood component is blood plasma or blood serum.

6. A method of preparing a bone mixture substantially as hereinbefore described.

7. Bone mixtures whenever prepared by the method according to claims 4 to 6.

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